Data sheet

SepFast™ Supor Q Column

SepFast Supor Q is designed for the capture of very large molecules (such as endotoxins, DNA, viruses and virus-like particles) with much increased binding capacity at high speed.

1. Properties

It is a strong anion exchange chromatography product supplied in a pre-packed ready-to-use column format. The working medium possesses a combination of small pores (50-100nm) and large pores (micro level). It shows fast accessibility to both small and large molecules. Therefore, it has comparable binding capacity to those of membrane or monolith products with respect to DNA or viruses. However, SepFast Supor Q shows much higher binding capacity to protein molecules than the membrane or monolith based products.

It shows the excellent binding capacity to both protein and macromolecules e.g. DNAs (see the Table 1) at high flow velocity. The capacity is further increased at lower flow velocity.

Table 1: A comparison of the dynamic binding capacity (DBC at 10% breakthrough) of different products (1 cm bed height with a cross-section area of 1 cm^2):

	SepFast Supor Q 10 CV	**Product A (particle format) 10 CV**	**Product A (particle format) 5 CV**	**Product B (particle format) 5 CV**
Calf thymus DNA fragment	**7075 μg/ml**	**105 μg/ml**	**1367 μg/ml**	**157 μg/ml**
Protein (BSA)	**133 mg/ml**	**84 mg/ml**	**116 mg/ml**	**1.7 mg/ml**

The base matrix is a composite of polysaccharides that have been highly cross-linked. The medium is stable in most of the chemical conditions experienced in the bioprocessing industry. The column design follows the same principle from small scale to large scale. The column material is disposable so it offers the benefit of cost reduction.

Table 2: Characteristics of SepFast Supor Q Columns:

Matrix	Highly cross-linked polysaccharide composites
Functional group	Strong anion Q, $-N^+(CH_3)_3$
Total ionic capacity	0.07 – 0.16 mmol Cl^-/ml medium
Particle size	40 μm (average)
Column material	Polypropylene (end-caps, meshes, stop plungers, column body for the 0.33 – 1 ml columns), acrylic (column body), NBR O-rings
Operational pressure	Up to 3 bar
Column pressure	4 bar
pH stability	2-14 (short term) and 2-12 (long term)
Working temperature	$+4^OC$ to $+30^OC$
Chemical stability	All commonly used buffers
Avoid	Alcohols (>20%), oxidizing agents, anionic detergents

2. Applications

SepFast Supor Q can be used in the final polishing step (shallow bed format such as 1-2 cm bed height) to remove trace amounts of impurities e.g. DNA, viruses, host cell proteins, endotoxins. The feedstock containing the target protein can pass through the column at a high speed (e.g. 10 packed column volumes) with much improved processing efficiency. This is usually not feasible with the conventional chromatography resins.

SepFast Supor Q can also be used for the binding-elution of both small (proteins) and large molecules (e.g. plasmid, virus, virus like particle) at short or deep bed format. Please contact us for further information.

3. Operations

The column is stored in 20% ethanol or in 0.02% sodium azide solution on delivery. It can be directly connect to a suitable chromatography system such as AKTA. Normally, the end with the product label should be connected as the top inlet. If there is no label difference between those two ends, the column can be connected either way.

The column need be equilibrated with at least 5 – 10 column volumes of the equilibration buffer and until the pH and conductivity signals become stable, before a sample is loaded.

The running pressure shall not exceed 3 bar during the operation.

After each application, seal the column ends and store it properly if re-use is expected.

4. Method optimization

We recommend scouting for optimal binding pH and for optimal ionic strength. SepFast Supor Q has a protein binding capacity (tested with BSA) almost independent of the flow velocity. Due to the fast pore accessibility, the binding step could be done at a faster flow velocity. We recommend special attention be paid to optimising elution conditions to avoid tailing in the elution step.

In general, balancing product recovery against process throughput is the major consideration when optimizing a method. However, for the purification of shearing-force sensitive molecules, the operational flow velocity needs to be optimised to balance the throughput and minimise the possible damage to the target molecule.

5. Process scaling up

SepFast Supor Q pre-packed columns range from small to large to support the scaling up of work. Please contact us for further information.

6. Maintenance

Depending on the individual applications, the column may be single use or re-used. For the re-use purpose, please see the following instructions.

Regeneration

After each run, elute any reversibly bound material either with a high ionic strength solution (e.g. 1M NaCl in buffer) or by increased pH.

Cleaning-in-place (CIP)

CIP is a procedure that removes strongly bound materials such as lipids, endotoxins and denatured proteins that remain in the column after regeneration. Regular CIP prevents the build up of contaminants in the packed bed and helps to maintain the column performance.

A specific CIP protocol should be developed for each process according to the type of contaminants present. The frequency of CIP depends on the nature of individual applications.

The following information works as a general guidance.

Salt of concentration up to 2 M can be used to clean the impurities bound by ionic interactions. The contaminants bound hydrophobically can be removed by using the following reagents: 1 M NaOH, low percentage non-ionic detergents (e.g. 0.1 – 2%), 30% isopropanol in basic or acidic conditions (e.g. in the presence of acetic acid or phosphoric acid). A combination of the above reagents can be explored as well. In general, the incubation time should be longer (e.g. from 30 minutes to 2 hours) to ensure full dissociation of the contaminants. Long contact time should be avoided when alcohols are used, as the acrylic column body may be damaged.

Sanitization
Sanitization using 0.5-1.0 M NaOH with a contact time of 1 hour is recommended.

7. Storage

The column should be equilibrated in binding buffer containing 20% ethanol or 0.02% sodium azide to prevent microbial growth. Store the column at a temperature of +4°C to +30°C. After storage, equilibrate the column with at least 5 bed volumes of running buffer before use.

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